FREE AMINO ACID AND RHEOLOGICAL MEASUREMENTS ON HYDROLYZED LACTOSE CHEDDAR CHEESE DURING RIPENING

- ABSTRACT -

Three lots each of conventional Cheddar and hydrolyzed lactose Cheddar cheese (HLCC) were analyzed for free amino acid content and examined for texture properties at 28-day intervals during an eightmonth ripening period. The average increases in total free amino acids in the control and the HLCC were from 500 µg/g in each cheese to 9,967 μ g/g and 14,564 μ g/g, respectively. Aspartic acid, proline and histidine were the only amino acids not significantly different between the two cheeses. Arginine, glycine, alanine, methionine and isoleucine increased slowly during ripening and accounted for 12% of the total free amino acids. Valine, tyrosine, phenylalanine, glutamic acid, leucine and lysine showed greater increases and accounted for about 80% of the total amount of free amino acids at all stages of ripening. Leucine increased the most, from 23.2 μ g/g to 1959.5 μ g/g in the control cheese and from 68.2 $\mu g/g$ to 2875.9 $\mu g/g$ in the HLCC. Arginine and methionine were the only two amino acids whose mole percentages did not significantly differ between the control and the HLCC. The mole percentages of aspartic acid, tyrosine, histidine, alanine, proline, lysine and glutamic acid decreased during the study in both the control and the HLCC. The mole percentage of leucine increased the most, from 5 to 15% in the control cheese and from 12% to 25% in the HLCC. The texture measurements for toughness, mechanical hysteresis, and modulus of elasticity showed significant (a0.05) treatment effects, but the changes during ripening were not significant. The toughness of the HLCC after eight months was 69% higher than in the control. Stiffness (modulus of elasticity) was 54% less and the damping capacity (mechanical hysteresis) was 93% more in the HLCC cheese than in the control. The differences in the texture measurements (toughness, mechanical hysteresis, and modulus of elasticity) on the controls and HLCC were greatest after 3 or 4 months and remained relatively constant thereafter. Therefore, proteolysis is related to texture and quality of cheese and accelerated ripening does seem to enhance desirable body and texture characteristics.

INTRODUCTION

CHEESE RIPENING is a complex process involving many physicochemical changes. Cheese ripening can be divided into two general phases, primary and secondary changes. Together they result in the accumulation of lactic acid, fatty acids and free amino acids. Secondary changes, specifically, are catalyzed by enzymes, primarily from microorganisms, that result in the formation of end products typical for each particular cheese variety or batch within a variety (Harper and Kristoffersen, 1956).

These chemical and physical changes cause the body of the freshly made cheese to lose its firm, tough, curdy texture and to become soft and mellow. This is related mainly to the progressive breakdown of the protein, which amounts to about 25% in Chedder cheese, to smaller polypeptides and gradual accumulation of amino acids. Free amino acids are also formed to some extent from nonprotein sources by transamination processes (Ernstrom and Wong, 1974).

This study was conducted to observe the differences during ripening of Cheddar cheese made by two processes: conven-

tional manufacture (the control) and manufacture from lactase-treated milk.

Hydrolyzed lactose Cheddar cheese (HLCC) manufacturing has only recently been reported (Thompson and Brower, 1974, 1976). Prehydrolysis of the milk's lactose accelerates cheese ripening with the result that after 3-4 months HLCC is very similar in all aspects to 6-, 7- or 8-month-old Cheddar made by conventional means. The underlying principle is the fact that lactase action will provide an immediate source of simple sugars leading to accelerated growth of the bacteria used in cheesemaking. Bacterial proteases, in turn, are greatly responsible for all succeeding effects. Earlier observations indicate that HLCC usually has better flavor, body and texture characteristics than comparable controls aged to their normal maturity (Thompson and Brower, 1976).

In order to study rate and extent of protein decomposition, a method was adopted to measure the gradually accumulating amino acids. Since there are so many intermediate and final compounds formed in maturing Cheddar cheese, it was necessary to choose compounds that could be easily identified and measured.

Very little has been reported on the development of free amino acids in Cheddar cheese during ripening. Most of the available data on this subject were generated in the early 1950's by less sensitive methods than are available today.

There appears to be no definite pattern among the basic free amino acids generated during ripening. Lysine appears at an early age (Bullock and Irvine, 1956). Histidine was not detected by Kosikowski (1951) and not until after 170 days by Bullock and Irvine (1956). Arginine was not detected until after 130 days of ripening (Bullock and Irvine, 1956). Lysine increased steadily during ripening (Bullock and Irvine, 1956; Kosikowski, 1951), but histidine and arginine stayed quite constant at relatively low concentrations (Bullock and Irvine, 1956). Another study revealed levels of arginine that continuously increased (Kosikowski, 1951). Arginine is of particular interest. Because of its repulsive, unpleasant, bitter-sweet taste, arginine has been held responsible for abnormal flavor development in cheese (Schormuller, 1968).

Several other free amino acids have been shown to continuously increase for about 1 yr before leveling off. These include alanine, aspartic acid, phenylalanine, serine and threonine (Bullock and Irvine, 1956). An exception was reported by Kosikowski (1951) who indicated only a small increase in threonine.

Glutamic acid, valine, methionine, isoleucine and leucine continuously increase in concentration throughout the ripening period (Bullock and Irvine, 1956; Kosikowski, 1951). Glutamic acid is found in the cheese initially (Kosikowski, 1951). Methionine is of special interest, since methional derived from it is considered the most important flavor compound in Cheddar cheese by Keeney and Day (1957).

Proline, glycine and tyrosine are the remaining free amino acids and do not have much in common with the others. Proline has been reported not to appear until after 180 days of ripening (Bullock and Irvine, 1956). It has also been reported

that proline is not present in ripened cheese at all (Kosikowski, 1951). Proline is the main flavor component in Swiss cheese (Harper and Kristoffersen, 1956). Glycine is found in all cheeses ripened more than 5 wk (Bullock and Irvine, 1956). It increases slowly during ripening with only a small amount present after 180 days (Kosikowski, 1951). Tyrosine is derived from the hydrolysis of peptides of low molecular weight (Schormüller, 1968). Its concentration in cheese does not increase after 180 days (Bullock and Irvine, 1956).

This study was conducted mainly to determine differences in free amino acid development between conventional Cheddar cheese and HLCC, but also to compare the data with previously reported work, and to investigate the texture properties of the cheeses, since rheological properties are dependent on proteolysis.

EXPERIMENTAL

Experimental design and statistical analysis

Six vats of cheese were manufactured: a control and a treatment (HLCC) each on three consecutive days. The only difference between control and treatment was the addition of 120g lactase (Maxalact, Food Grade, 40,000 ONPH/g; Enzyme Development Corp., New York, NY) to the 360 kg milk in the treatment vat. The cheese from each vat was heat-sealed into four 9.1-kg plastic-covered blocks (Parakote Plain No. 20, American Can Co., Greenwich, CT). One block, selected as an experimental unit, was subdivided by drawing 2-inch-squares on the surface of the plastic coating. Each square was randomly assigned a sampling date. Two randomly selected squares were used for each sampling date. Samples were removed with a cheese trier initially and eight times thereafter at 28-day intervals. All samples were kept frozen in sealed plastic bags until used in analysis.

This experiment was a three-factor factorial design with treatment, day of manufacture and month of sampling as factors. All samples were subsampled for free amino acid analysis, resulting in four repetitions. An analysis of variance was conducted using the average values of the repetitions; this analysis determined whether the treatment effect was significant. For the Duncan mean separation of the data by months, a separate analysis of variance was conducted for the control and the treatment. The appropriate mean squares were then used for mean separation. The statistical analyses were done with library programs on file at the University's Computation Center.

Cheddar cheese cultures

All Cheddar cheeses were made with Hansen's Dri Vac lactic starter culture (Chr. Hansen's Laboratory, Inc., Milwaukee, WI) which was subcultured in Matrix Cultura Media (Galloway-West Co., Fond du Lac, WI) according to directions. Addition to the cheese milk was at a rate of 1% active culture.

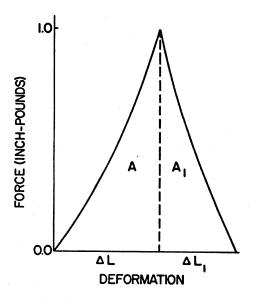


Fig. 1—Typical force-deformation curve from the Instron Universal Testing Machine.

Cheddar cheese manufacture

Two vats with 360 kg milk each were used on three successive days. Each day pasteurized milk was standardized to 3.7% fat and heated to 31°C. Lactase (120g) was added to the treatment vat. The milk was held at this temperature for 1 hr and then the starter was added to each vat. After another hour at 31°C the coagulant was added. This consisted of 90 ml rennet (Chr. Hansen's Laboratory, Inc., Milwaukee, WI) diluted with about 1500 ml water. The cheese was ready for cutting in about 30 min. After cutting, the curd was allowed to remain quiescent for 10 min, then cooking was initiated together with gentle stirring. The vat contents were brought to 32.5°C in 10 min. An additional 20 min of cooking and stirring was used to raise the temperature to 38°C. At this temperature, the cheese was "cooked" for 30-35 min so the curd would attain the proper degree of firmness. The whey was then drained. the curd packed along the vat sides and cut into slabs after 10 min. During the cheddaring process these slabs were turned every 15 min. The whey acidity was periodically titrated and upon reaching 0.50% (expressed as lactic acid) the slabs were milled. Salting was done with 1.1 kg of NaCl in three intervals 5 min apart The milled curd was then packed in 9.1 kg metal curd boxes and pressed overnight at a pressure of 60 psig (4.22 kg/cm²). The blocks were wrapped, sealed and placed in a curing room kept at 9°C.

Free amino acid analysis

Samples weighing 0.25-2.0g, depending on the cheese's age, were placed in the cup of a Sorvall Omni Mixer (Ivan Sorvall, Inc., Norwalk, CT). To this were added 25 ml of an aqueous solution of 1% picric acid and 250 μ l of an internal standard containing 5 μ mole of norleucine (J.T. Baker Chemical Co., Phillipsburg, NJ) and 5 μ mole of S- β -(4-pyridylethyl)-DL-penicillamine (4-PEP) (Pierce Chemical Co., Rockford, IL) for the long and short columns, respectively. Each sample was blended for 2 min, then centrifuged for 10 min at 3000 rpm. The supernatant was poured through a prepared resin bed (10 cm, Dowex 2X8-200 (CL)). The remaining sample was washed twice with 10 ml of 0.02N HCl. The washings were also added to the column. The eluate was collected in a 50-ml volumetric flask and brought to volume with 0.02N HCl. These samples were then frozen. At a later date, 1-ml quantities were analyzed on a Beckman Model 120 C amino acid analyzer (Beckman Instruments, Inc., Palo Alto, CA).

The resin beds were prepared by first washing the resin with 1N HCl, decanting the fines after the resin settled. The resin was then poured into 50-ml burets, rinsed with 1N HCl, allowed to pack and washed with two 20-ml portions of distilled water.

Rheology measurements

Mechanical measurements were made with an Instron Universal Testing Machine. A full-scale deflection on the chart was 1 lb and all samples had a 1-lb force applied before unloading. The crosshead and chart speeds were 0.5 inch and 5 inch/min, respectively. Cheese samples to be tested were cylinders 11/16 inch long with a diameter of 7/16 inch $(17.5 \times 11.1 \text{ mm})$.

The following rheological evaluations were made: (a) toughness, the work required to cause rupture in the cheese; (b) hysteresis, the energy absorbed by a material in a cycle of loading and unloading or, the damping capacity, (c) modulus or stiffness, the ratio of stress to corresponding strain below the proportional limit, and (d) degree of elasticity, the ratio of elastic deformation to the sum of elastic and plastic deformation when a material is loaded to a certain load and then unloaded to zero load. The force-deformation curve obtained was used to derive values for the various rheological measurements. A typical curve is shown in Figure 1.

Toughness was calculated by measuring the area under the force-deformation curve to the left of a perpendicular line drawn to the abscissa from the point where the curve attained a negative slope. This is shown as area A in Figure 1. Hysteresis is the difference between area A and the other point of the curve resulting from unloading (area A_1). Modulus (stiffness) was calculated by the following formula: Elasticity = $(F/A_2)/\Delta L/L$), where F = force applied, A_2 = cross-sectional area of the cylinder of cheese, ΔL = length as shown in Figure 1 for the force-deformation curve to peak and L = length of cylinder. The degree of elasticity is the ratio of ΔL_1 and ΔL .

RESULTS & DISCUSSION

A SUMMARY of the mole percentages of the 14 amino acids monitored during the ripening of the control cheese and HLCC is contained in Tables 1 and 2. Arginine and methionine were the only amino acids whose mole percentages were not signifi-

Table 1-Mole percentages of 14 free amino acids during ripening of conventionally made Cheddar cheesea

Amino acid	April	May	June	July	August	September	October	November	December
Lysine	22.8a	18.8b	17.5bc	18.9ab	15.2bc	16.3bc	15.2bc	15.5bc	14.0c
Histidine	4.9a	2.7b	1.7c	1.7c	1.0d	1.7c	1.7c	1.3cd	1.8c
Arginine	0.7a	1.3ab	1.2ab	2.3ab	2.6ab	2.8ab	2.3ab	2.2ab	3.8b
Aspartic acid	7.0a	5.9ab	5.2bc	4.9bc	4.6c	4.7bc	4.9bc	4.4c	4.2c
Glutamic acid	20.5a	20.0a	18.0bc	16.2d	18.4b	16.8cd	16.1d	18.0bc	17.0cd
Proline	15.3a	8.9b	6.3bc	6.1c	4.1cd	6.3bc	5.3cd	3.3d	4.0cd
Glycine	0.9a	1.9bc	1.8b	2.5d	2.2cd	3.0e	3.1ef	3.2ef	3.5f
Alanine	7.0a	7.0a	5.7b	4.9bc	5.1bc	4.5bc	4.7bc	4.8bc	4.3c
Valine	4.0a	5.5b	6.7c	7.6de	7.1cd	7.7de	8.8f	8.3ef	8.2ef
Methionine	0.7a	2.7d	4.9b	4.1bc	3.3cd	3.1d	3.5cd	3.1d	2.9d
Isoleucine	0.5a	1,0b	1.5c	1.5c	2.1d	2.2de	2.6e	2.1d	2.3de
Leucine	4.9a	13.1b	16.3c	17.3cd	20.5e	19.1de	19.4de	21.3e	20.0e
Tyrosine	6.0a	4.1b	4.1b	2.8c	3.1bc	3.0bc	3.2bc	3.2bc	3.7bc
Phenylalanine	4.8a	7.8b	9.4cd	9.3cd	10.4d	9.2c	9.1c	9.2c	8.3bc

a Within rows means followed by the same letter are not significantly different ($\alpha 0.05$) from each other.

cantly different $(\alpha 0.05)$ between control and treatment throughout ripening.

The amounts of aspartic acid, tyrosine, histidine, alanine, proline, lysine and glutamic acid, as mole percentages, decreased from their initial values during the ripening study, while valine, isoleucine, glycine, leucine and phenylalanine increased throughout ripening.

The extent of proteolysis during cheese ripening was ascertained by measuring the concentration of free amino acids produced (Fig. 2). The difference between the control and HLCC became evident very early. Both contained an initial level of about 500 μ g of free amino acids per gram of cheese. Total free amino acids in the control cheeses increased to 9,967 μ g/g and in the HLCC to 14,654 μ g/g of total free amino acids after 9 months. That constitutes a 47% higher level of free amino acids in the HLCC than in the control cheeses.

A summary of the microgram data for the individual amino acids, together with the Duncan mean separation results, is contained in Table 3 for the control Cheddar cheese and Table 4 for the HLCC. The data reported are averages of the three replications of the experiment.

Many of the earlier investigators were unable to detect certain amino acids early in the ripening period because of insensitivity of the methods used.

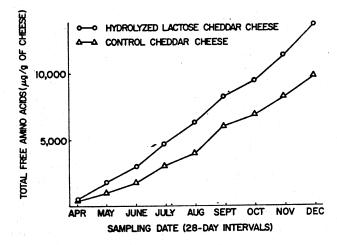


Fig. 2—Changes in total free amino acids during ripening of conventional and hydrolyzed lactose Cheddar cheeses.

The changes in the amounts of free amino acids per gram of cheese for aspartic acid, proline and histidine were the only ones for which the treatment effect was not significant. Free aspartic acid in the control cheese increased from $32.7 \,\mu\text{g/g}$ to

Table 2-Mole percentages of 14 free amino acids during ripening of hydrolyzed lactose Cheddar cheesea

Amino acid	April	May	June	July	August	September	October	November	December
Lysine	16.9a	14.6bc	15.9ab	13.2cde	14.6bcd	12.5e	12.5e	12.9de	12.6e
•	3.6a	1.5b	1.0c	1.0c	1.1c	1.1c	1.2bc	1.2bc	1.3bc
Histidine		1.1bc	1.5abc	2.6bcd	2.5bcd	2.9cd	2.9cd	3.3d	2.9cd
Arginine	0.5a	4.0b	3.2de	3.1e	3.3cde	3.6bcd	4.0b	3.4cde	3.7bc
Aspartic acid	7.0a			15.8abc	15.5c	15.9abc	17.0abc	16.9abc	17.7ab
Glutamic acid	17.9a	15.8bc	15.4c		3.5bcd	4,9b	3.8bcd	3.8bcd	3.8bcd
Proline	10.7a	4.7bc	3.3cd	3.0d		3.2b	3.4b	3.2b	4.0c
Glycine	1.6a	2.0a	2.0a	2.1a	2.9b		4.2b	4.1b	4.3b
Alanine	5.8a	4.7ab	4.2b	3.9b	4.2b	4.2b		9.0cde	9.2e
Valine	5.9a	7.4b	7.7b	8.5cd	8.4c	9.0de	9.0cde		3.1c
Methionine	1.1a	3.9b	3.1c	3.2c	3.0c	3.1c	3.2c	3.0c	
Isoleucine	1.3a	2.3cd	2.0b	2.0bc	2.4de	2.6fg	2.8fg	2.8fg	2.99
Leucine	12.4a	20.1b	23.2cd	24.7d	24.0d	23.6d	24.0d	23.6d	21.6bc
Tyrosine	5.9a	4.7b	3.9bc	4.3bc	3.7c	4.0bc	4.0bc	4.1bc	4.5b
Phenylalanine	9.2c	13.1a	13.5a	12.6a	10.8b	9.8bc	9.4c	9.0cd	7.6d

 $^{^{}f a}$ Within rows means followed by the same letter are not significantly different (lpha0.05) from each other.

Table 3—Free amino acids (mg/g of cheese) during the ripening of conventional Cheddar cheese*

Amino acid	April	May	June	July	August	September	October	November	Decembe
Lysine	116.1	205.4a	350.3ab	658.8b	671.5b	1113.8c	1142.5c	1389.2cd	
Histidine	26.8a	30.3a	33.9a	56.4a	46.1a	119.8b	125.4b	120.2b	1553.7d
Arginine	4.3a	9.4ab	23.3ab	76.7ab	102.6abc	173.1abc	180.7bc	260.9cd	202.8c
Aspartic acid	32.7a	57.7a	87.9ab	138.3bc	165.6c	267.0d	313.8de	352.1e	410.9d
Glumatic acid	105.0a	218.6ab	355.9ab	541.3bc	778.1cd	1116.5de	1210.8e	1604.1f	427.71
Proline	60.9	73.7ab	90.2ab	142.7abc	136.3abc	341.2d	290.2cd		1822.3f
Glycine	2.3a	10.6ab	19.2ab	42.5ab	48.5b	105.0c	120.0c	227.7bcd	339.4d
Alanine	21.6a	46.3ab	67.3bc	97.0cd	126.6de	170.2ef	210.0f	144.1c	189.8d
Valine	16.5a	48.4a	104.4ab	196.5b	237.3b	421.0c	517.0cd	257.2g	290.19
Methionine	3.4a	30.1a	94.0b	128.7b	139.5b	204.3c		586.0d	741.6e
soleucine	2.3a	10.5a	26.9a	43.4ab	71.4b	123.6c	243.5cd	280.0d	339.7e
_eucine	23.2a	129.8a	292.2ab	523.1bc	774.0cd		184.6d	167.1d	238.2e
Tyrosine	38.1a	55.3a	75.4ab	117.0ab		1099.7de	1289.1e	1700.3f	1959.41
Phenylalanine	26.9a	97.1ab	209.bc		157.0b	262.4c	295.6c	358.0c	455.5d
,		37.1ab	209.DC	346.3cd	480.3d	641.8e	755.3e	918.3f	1042.2f

^a Within rows means followed by the same letter are not significantly different (α 0.05) from each other.

427.7 μ g/g in the hydrolyzed lactose cheese. Proline (control, 60.9-339.4; HLCC, 50.8-442.8) and histidine (control, 26.8-202.8; HLCC, 23.0-205.4) had similar patterns of development in both cheeses; These three amino acids accounted for about 8% of the total free amino acids found and were characterized by only slight changes, less than 100 μ g/g, during the first 120 days of ripening.

Arginine, glycine, alanine, methionine and isoleucine accounted for 12% of the total quantity of the free amino acids. All increased significantly due to treatment effects. Arginine (control, 4.3-410.9; HLCC, 3.5-506.9) steadily increased, as had been pointed out earlier by Kosikowski (1951). No leveling off was found in either cheese, although it has been anticipated (Bullock and Irvine, 1956). It is of interest to note that the final concentration of arginine is higher in the HLCC than in the control and that arginine has been reported to be responsible for an unpleasant or bitter-sweet taste (Schormüller, 1968). However, no bitterness was observed in the cheese. Potential for the defect does exist. Glycine (control, 2.3-189.8; HLCC, 5.0-316.6) initially increased slowly during ripening, but after 90 days its concentration increased rapidly. This development is different from that observed by Kosikowski (1951) who stated that only small amounts were present after 180 days. Alanine (control, 21.6-290.1; HLCC, 21.9-396.5) continually increased during the study and

reached a constant value near the end as previously reported for Cheddar cheese (Bullock and Irvine, 1956). Methionine (control, 3.4–339.7; HLCC, 6.8–480.3) and isoleucine (control, 2.3–238.2; HLCC, 7.0–403.6) also continually increased throughout the study. This is in agreement with earlier work on Cheddar cheese (Bullock and Irvine, 1956; Kosikowski, 1951). It is important to note the larger amount of methionine in the HLCC since methional, derived from methionine, is considered by Keeney and Day (1957) the most important flavor compound in Cheddar cheese.

Valine, tyrosine, phenylalanine, leucine and lysine, accounting for 80% of the total, increased the most. Valine (control, 16.5-741.6; HLCC, 29.0-1110.0) increased greatly during the ripening period. Valine and leucine (control, 23.2-1959.4; HLCC, 68.2-2875.9) are reported in the literature as continually increasing throughout Cheddar cheese ripening (Bullock and Irvine, 1956; Kosikowski, 1951), as was observed also in this study. Leucine displayed the largest increase and also the largest difference between control and HLCC. The other free amino acids also increased steadily: tyrosine (control, 38.1-455.5; HLCC 45.2-890.7), phenylalanine (control, 26.9-1042.2; HLCC, 63.8-1399.4), glutamic acid (control, 116.1-1553.7; HLCC, 102.9-2065.3).

The mechanical measurements for toughness, hysteresis, and modulus of elasticity showed significant ($\alpha 0.05$) treatment

Table 4—Free amino acids (mg/g of cheese) during the ripening of hydrolyzed lactose Cheddar cheese®

Amino acid	April	May	June	July	August	September	October	November	December
Lysine	102.9a	282.4ab	504.6abc	659.1bcd	978.2cde	1118.9de	1220.0-4	4700.46	2005 2-
Hisitidine	23.0a	28.3a	33.4ab	51.2ab	76.2bc		1280.9ef	1723.1fg	2065.33
Arginine	3.5a	21.9a	48.8ac	130.2acd	163.7acd	99.8cd	128.9de	1681.3eg	205.4f
Aspartic acid	39.1a	66.0ab	88.9ab	135.9ab		262.6cde	302.9def	417.5ef	506.9f
Glutamic acid	109.9a	294.7ab	482.4ab		199.8b	380.5c	358.8cd	417.4cd	517.6cd
Proline	50.8a	66.7a		788.8bc	1035.9cd	1405.5de	1711.2e	2257.3f	2559.0f
Glycine	5.0a		77.3a	114.5ab	193.4bc	323.6d	292.1cd	392.8de	442.8e
Alanine		19.6a	32.1ab	54.5ab	99.8bc	145.2cd	172.7de	220.1e	316.6f
Valine	21.9a	53.9ab	78.5ab	121.5bc	168.8cd	223.8de	257.8e	336.6f	396.5f
	29.0a	111.0a	191.5ab	336.8bc	450.7cd	608.7de	723.6e	952.6f	1110.0f
Methionine	6.8a	78.6ab	97.4bc	157.7cd	199.4de	269.aef	327.af	408.8g	480.39
Isoleucine	7.0a	39.0a	55.2ab	90.3ab	143.7bc	204.7cd	256.8de	338,2ef	403.6f
Leucine	68.2a	342.2ab	653.3bc	1102.6cd	1425.0de	1868.1ef	2162.9f		
Tyrosine	45.2a	109.5ab	151.8ab	267.2abc	302.4bc	432.8cd		2803.2g	28.75.99
Phenylaline	63.8a	271.7b	458.6c	674.3d	771.7e	432.8cg 937.1f	507.9cd 1048.6g	67.65de 1186.7h	890.7e 1399.4e

 $^{^{}a}$ Within rows means followed by the same letter are not significantly different (lpha 0.05) from each other .

Table 5—Rheological measurements of toughness, hysteresis, modulus of elasticity and degree or elasticity throughout the ripening period of conventional (Con) and hydrolyzed lactose Cheddar cheeses (HLCC)

	Toughness (in-lbs)			Hysteresis (in-lbs)			Modulus (lb-in²)			Degree elasticity (%)	
Month	Conv	HLCC	Diffa (%)	Conv	HLCC	Diffa (%)	Conv	HLCC	Diffa (%)	Conv	HLCC
		0.000	10.3	0.037	0.041	10.8	39.2	33.0	-15.8	50.6	53.5
Vlay	0.057	0.068	19.3		0.044	41.9	41.3	31.4	-24.0	51.5	54.7
lune	0.049	0.067	36.7	0.031	• • • • •		46.4	27.7	-40.3	54.6	52.5
July	0.042	0.075	79.0	0.023	0.047	104.3	· - · .				52.7
	0.042	0.086	104.7	0.020	0.061	205.1	51.4	28.0	-45.5	59.2	
August				0.027	0.054	100.0	59.1	31.7	-46.4	54.4	49.4
September	0.040	0.077	92.5		•		54.1	34.7	-35.9	51.9	47.4
October	0.041	0.077	87.8	0.027	0.055	103.7		- :	-26.0	51.8	49.8
November	0.051	0.083	63.7	0.032	0.060	89.5	44.0	32.6		31.0	

The percent difference is the treatment minus the control expressed as a pecentage of the control.

effects, but the changes during ripening were not significant (Table 5). The toughness of the HLCC cheese after eight months was 64% higher than in the control. Toughness was correlated to the modulus of elasticity and hysteresis measurement during ripening by -0.89 and 0.99, respectively. The modulus of elasticity or stiffness was 26% less and the hysteresis or damping capacity was 90% more in the HLCC than in the control. The degree of elasticity for HLCC was 50% and for the control 52%, a nonsignificant difference.

The percent differences in texture measurements between controls and HLCC for toughness, hysteresis, and modulus reached maximum values after 3 or 4 months and remained relatively constant throughout the remainder of the ripening period (Table 5).

The rheological data indicate that, overall, HLCC arrives at the same desirable body and texture characteristics several months earlier than conventionally made Cheddar cheese.

REFERENCES

Bullock, D.H. and Irvine, O.R. 1956. A chromatographic study of Cheddar cheese ripening. J. Dairy Sci. 39: 1229.

Ernstrom, C.A. and Wong, N.P. 1974. In "Fundamentals of Dairy Chemistry," Ed. Webb, G.H., Johnson, A.H. and Alford, J.A. Avi Publishing Company, Inc., Westport, CT.
Harper, W.J. and Kristoffersen, T. 1956. Biochemical aspects of cheese ripening. J. Dairy Sci. 339: 1773.
Keeney, M. and Day, E.A. 1957. Probable role of the Streekey degrada.

Keeney, M. and Day, E.A. 1957. Probable role of the Strecker degradation of amino acids in development of cheese flavor. J. Dairy Sci. 40: 874.

Kosikowski, F.V. 1951. The liberation of free amino acids in raw and pasteurized milk Cheddar cheese during ripening. J. Dairy Sci. 34: 235.

Schormuller, J. 1968. The chemistry and biochemistry of cheese ripening. Adv. Food Res. 11(1): 231.

Thompson, M.P. and Brower, D.P. 1974. Manufacture of Cheddar cheese from hydrolyzed lactose milks. J. Dairy Sci. 57: 598. Thompson, M.P. and Brower, D.P. 1976. Hydrolyzed lactose cultured

dairy products. 1. Manufacture of Cheddar cheese. Cultured Dairy Products J. 11(1): 22.

Ms received 4/28/77; revised 7/9/77; accepted 7/10/77.

Presented at the 37th Annual Meeting of the Institute of Food Technologists, Philadelphia, PA, June 5-8, 1977.

Paper No. 5262 in the Journal Series of the Pennsylvania Agricul-

tural Experiment Station.

The authors express appreciation to Nancy P. Warner for her technical assistance and Dr. C.T. Morrow for help with the rheological measurements. This study was supported, in part, by Dairy Research Inc., Rosemont, IL.

Ciegler, A., Vesonder, R.F. and Jackson, L.K. 1977. Production and biological activity of patulin and citrinin from Penicillium expansum. Appl. Env. Microbiol. 33: 1004.

Damodaran, C., Ramadoss, S.S. and Shanmugasundaram, E.R.B. 1973.

A rapid procedure for the isolation, identification and estimation of

citrinin, Anal. Biochem. 52: 482.
Davis, N.D., Dalby, D.K., Diener, U.L. and Sansing, G.A. 1975. Medium-scale production of citrinin by Penicillium citrinum in a semi-synthetic medium. Appl. Microbiol. 29: 118.

Diener, U.L. 1960. The mycoflora of peanuts in storage. Phytopath-

ology. 50: 220.
Früs, P., Hasselager, E. and Krogh, P. 1969. Isolation of citrinin and oxalic acid from Penicillium viridicatum Westling and their nephrotoxicity in rats and pigs. Acta Path. Microbiol. Scand. 77: 559.
Hald, B. and Krogh, P. 1973. Analysis and chemical confirmation of citrinin in barley. J. Assoc. Offic. Anal. Chem. 56: 1440.
Hueh, D.P.H. 1973. Inhibition of aflatoxin biosynthesis by dichlorvos. J. Agric. Food Chem. 21: 468.
Krogh, P. Hosselager, F. and Friis, P. 1970. Studies on fungal nephro-

Krogh, P., Hasselager, E. and Friis, P. 1970. Studies on fungal nephrotoxicity, Acta Path. Microbiol. Scand. Sec. B 78: 401.
 Krogh, P., Hald, B. and Pedersen, E.J. 1973. Occurrence of ochratoxin

A and citrinin in cereals associated with mycotoxic porcine nephropathy. Acta Path. Microbiol. Scand. Sec. B. 81: 689.

Mislivec, P. and Tuite, J. 1970. Species of Penicillium occurring in freship. freshly harvested dent corn kernels. Mycologia 62: 67.

Narasimhan, K.S. and Rangaswami, G. 1968. Effect of some fumigants on the microflora of sorghum seeds. Indian Phytopathol. Soc. Bull. Raghunathan, A.N., Murhu, M. and Majumder, S.K. 1969. Control of internal fungi of sorghum by fumigation. J. Stored Prod. Res. 5:

389 Rao, H.R.G. and Harein, P.K. 1972. Dichlorvos as an inhibitor of aflatoxin production on wheat, corn, rice and peanuts. J. Econ. Entomol. 65: 988.

Entomol. 60: 986.

Saito, M., Enomoto, M. and Tatsuno, T. 1971. Yellowed rice toxinscitrinin. In "Microbiol Toxins," Vol. 6, Ed. Ciegler, A., Kadis, S. and Ail, S.J. p. 357. Academic Press, Inc., New York.

Scott, P.M., van Walbeek, V., Kennedy, B. and Anyeti, D. 1972. Mycotoxins (ochratoxin A, citrinin, and sterigmatocystin) and toxigenic toxins and other estimators.

fungi in grains and other agricultural products. J. Agric. Food Chem.

20: 1103.
Vandegraft, E.E., Shotwell, O.L., Smith, M.L. and Hesseltine, C.W. 1973. Mycotoxin production affected by insecticide treatment of wheat. Cereal Chem. 50: 264.
Wu, M.T. and Ayres, J.C. 1974. Effects of dichlorvos on ochratoxin production by Aspergillus ochraceus. J. Agric. Food Chem. 22: 536.
Wu, M.T., Ayres, J.C. and Koehler, P.E. 1973. Production of citrinin by Papiallium of country-guired ham. Appl. Microbiol. 27. Penicillium viridicatum on country-cured ham. Appl. Microbiol. 27:

Yao, R.C. and Hsieh, D.P.H. 1974. Step of dichlorvos inhibition in the pathway of aflatoxin biosynthesis. Appl. Microbiol. 28: 52. Ms received 6/21/77; revised 9/30/77; accepted 10/5/77.

Presented at the 37th Annual Meeting of the Institute of Food Technologists, Philadelphia, PA. June 5-8, 1978. This investigation was supported by Grant No. 5-R01-FD00155-10 from the Food & Drug Administration.